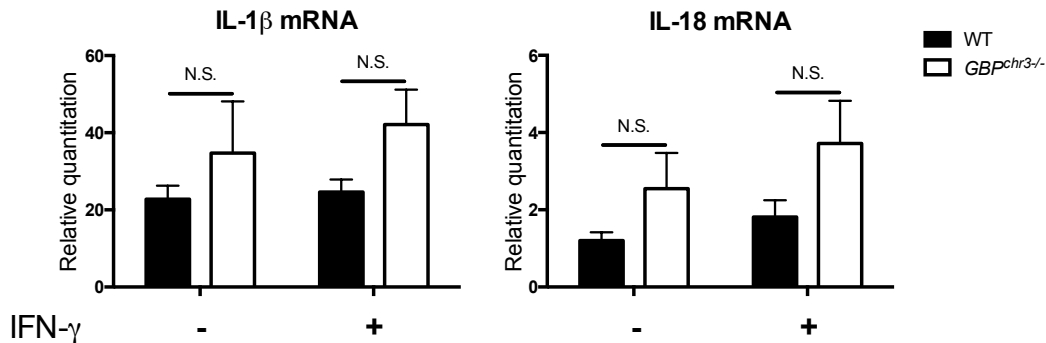
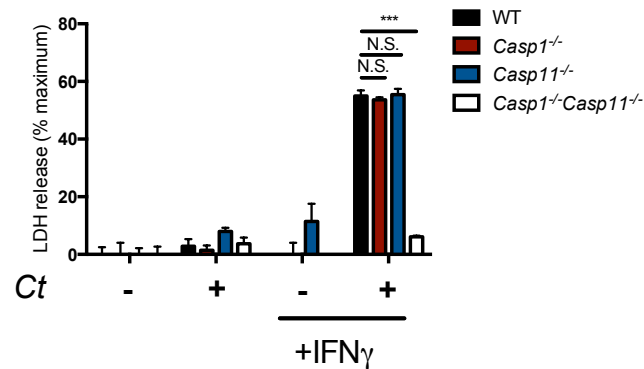


**Figure S1. GBP2 fails to localize to *C. muridarum* inclusions in IFN $\gamma$ -primed macrophages**

IFN $\gamma$ -primed wildtype and GBP<sup>chr3-/-</sup> BMMs were infected with *C. muridarum* at an MOI of 3 and stained with Hoechst, anti-Chlamydia LPS and anti-GBP2 antibodies. Arrow points at GBP2-positive vesicles found in wildtype cells.

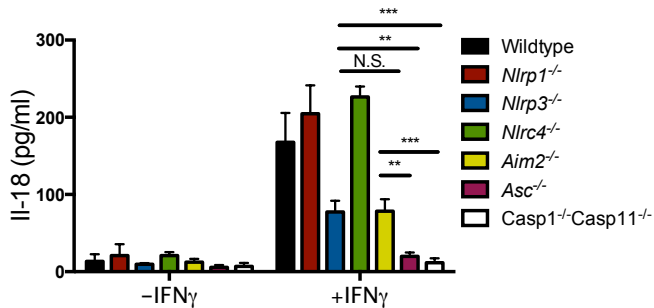


**Figure S2. IL-1 $\beta$  and IL-18 transcription is unchanged in GBP-deficient macrophages**  
Wildtype and GBP<sup>chr3-/-</sup> BMMs were treated overnight with 100 U/ml IFN $\gamma$  or left unprimed. Cells were then infected with *C. muridarum* at an MOI of 30. RNA was extracted at 4 hpi and relative IL-1 $\beta$  and IL-18 transcript levels were determined via qPCR. Data represents transcript levels relative unprimed, uninfected wildtype control. Data are shown as mean  $\pm$  SEM of 3 independent experiments. Two-way ANOVA was used to calculate statistical significance with \* $p < 0.05$ ; N.S. = not significant.



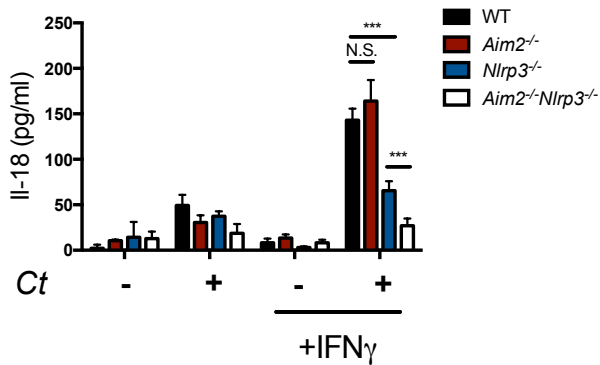
### Figure S3. *C. trachomatis* infections activate canonical and noncanonical inflammasomes

Wildtype, *Casp1*<sup>-/-</sup>, *Casp11*<sup>-/-</sup> and *Casp1*<sup>-/-</sup>*Casp11*<sup>-/-</sup> BMMs were primed with 100 U/ml IFN $\gamma$  or left unprimed and subsequently infected with *C. trachomatis* at an MOI of 30. LDH release was measured at 8 hpi. Statistical significance was calculated using two-way ANOVA. \*\*\* $p < 0.001$  and N.S. = not significant.



### Figure S4. *C. muridarum* infections activates AIM2 and NLRP3 inflammasomes

Wildtype, *Nlrp1*<sup>-/-</sup>, *Nlrp3*<sup>-/-</sup>, *Nlrc4*<sup>-/-</sup>, *Aim2*<sup>-/-</sup>, *Asc*<sup>-/-</sup> and *Casp1*<sup>-/-</sup>/*Casp11*<sup>-/-</sup> BMMs were primed with 100 U/ml IFN $\gamma$  or left unprimed and infected with *C. muridarum* at an MOI of 30. IL-18 secretion was measured via ELISA at 24 hpi. Data are shown as mean  $\pm$  SD of 3 independent wells and are representative of 3 experiments. Statistical significance was calculated using two-way ANOVA. \*\*p < 0.01, \*\*\*p < 0.001 and N.S = not significant.



### Figure S5. *C. trachomatis* infections activate AIM2 and NLRP3 inflammasomes

Wildtype, *Aim2*<sup>-/-</sup>, *Nlrp3*<sup>-/-</sup> and *Aim2*<sup>-/-</sup>*Nlrp3*<sup>-/-</sup> BMMs were primed with 100 U/ml IFN $\gamma$  or left unprimed. Cells were then infected with *C. trachomatis* and supernatants collected at 24 hpi. IL-18 concentrations in cell supernatans were determined by ELISA. Data are depicted as mean  $\pm$  SD of 3 independent wells and are representative of 3 experiments. Statistical significance was calculated using two-way ANOVA. \*\*\*p < 0.001 and N.S = not significant.